

Applicant : George B. Witman et al.  
Serial No. : 09/866,582  
Filed : May 24, 2001  
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Attorney's Docket No.: 07917-145001 / UMMC 01-23

**REMARKS**

The amendments to the specification add no new matter.

**Restriction Requirement**

Responsive to the action mailed June 7, 2002, applicant elects the invention of Group I (claims 1-7 and 18-23). The election is made without traverse.

**Sequence Compliance**

Applicants submit herewith a Sequence Listing in computer-readable form as required by 37 CFR §1.824. In addition, applicants submit a substitute Sequence Listing as required under 37 CFR §1.823(a) and a statement under 37 CFR §1.821(f).

Applicants hereby submit that the enclosures fulfill the requirements under 37 C.F.R. §1.821-1.825. The amendments in the specification merely insert sequence identifiers in the specification and replace the original Sequence Listing with a substitute Sequence Listing. No new matter has been added.

**CONCLUSION**

Applicants request that the claims be examined. Attached hereto is a marked-up version of the changes made to the specification by the current amendment.

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Please apply any charges or credits to Deposit Account No. 06-1050, referencing attorney docket number 07917-145001.

Respectfully submitted,

Date: December 6, 2002

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**"Version With Markings to Show Changes Made"**

In the specification:

Paragraph beginning at page 78, line 21, has been amended as follows:

Portions of the IFT88 peptide sequence (LEGETDQA (SEQ ID NO:43) and GIDPYCVE (SEQ ID NO:44)) were used to design two degenerate oligonucleotide PCR primers (GA[A/G] AC[C/G/T] GA[C/T] CA[A/G] GC[C/G/T] GA[C/T] AA[A/G] TA (SEQ ID NO:45) and GC [C/T]TC [A/C/G]AC [A/G]CA [A/G]TA [A/C/G]GG [A/G]TC [A/G]AT (SEQ ID NO:46)). These primers amplified a 365-bp fragment of genomic DNA that contained parts of two exons and a 132-bp intron. This fragment of genomic DNA was used to screen a *Chlamydomonas* cDNA library made from cells undergoing division (Pazour and Witman, unpublished). Two positive clones were identified and sequenced by primer walking. These two clones were similar except for the sequences at their 5'-ends. IFT88cDNA-1 was longer than IFT88cDNA-2 and appeared to have a short region of poly-A inappropriately fused to the 5'-end, probably the result of a cloning artifact. One *Chlamydomonas* IFT88 EST clone is in Genbank (accession number AV395576). This EST sequence, which is from the 5' end of the gene and overlaps the cDNA clones, was used to define the 5'-end of the cDNA sequence.

Paragraph beginning at page 81, line 6, has been amended as follows:

In order to learn more about the structure and function of the proteins that make up the IFT particle, we cloned and sequenced the IFT88 protein, formerly known as p88 (Cole et al., 1998). To do this, *Chlamydomonas* IFT particles were purified from the matrix of isolated flagella by sucrose density gradient centrifugation and two-dimensional gel electrophoresis. IFT88 was cleaved by trypsin and two internal peptides were microsequenced (Cole et al., 1998), yielding the sequences AATNLAFLYFLEGETDQADKYSEMALK (SEQ ID NO:47) and SLFNEAAGIDPYCVEAIYNLGLVSQR (SEQ ID NO:48). Degenerate PCR primers were designed from these sequences and used to amplify a fragment of genomic DNA. A cDNA library was screened with the genomic fragment and the resulting clones were sequenced by

primer walking. Southern blots indicated that there is only one copy of the *IFT88* gene in the *Chlamydomonas* genome.

Paragraph beginning at page 89, line 12, has been amended as follows:

**Cloning IFT20:** *Chlamydomonas* IFT20 was purified and the sequence of two tryptic peptides was obtained (GVYFDEDFHVR (SEQ ID NO:49) and YVSAIDQQVER (SEQ ID NO:50)) (Cole et al., *J. Cell Biol.* 141:993-1008, 1998). A degenerate PCR primer designed from the first peptide sequence was used in combination with an oligo-dT primer to amplify most of the coding sequence from reverse-transcribed cDNA. The remainder of the gene was amplified from a *Chlamydomonas* cDNA library in lambda ZapII (Stratagene) with a vector primer (M13Rev) and a IFT20-specific primer designed from the sequence of the first PCR product. The open reading frame contained within these clones encodes a 15.6-kD peptide containing both tryptic peptides.